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MANUNEMA PECTENOPHORA SP. NOV. (PERESIANIDAE, LEPTOLAIMINA), A NEMATODE POSSESSING UNUSUAL MALE SUPPLEMENTARY ORGANS

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Summary

STEWART, A. C. & NICHOLAS, W. L. (1995) *Manunema pectenophora*, sp. nov. (Peresianidae, Leptolaimina), a nematode possessing unusual male supplementary organs. *Trans. R. Soc. S. Aust.* **119**(4), 163-169, 30 November, 1995.

Manunema pectenophora, sp. nov., with three unique pre-anal male supplementary organs, is described. These are comb-like organs held clear of the body on short rods. Two previously-described species of *Manunema*, the sole genus in the Peresianidae, possess tubular supplements. *M. pectenophora* also differs from the other species in that the single testis is anterior. All *Manunema* species possess four long cephalic setae, no labial setae or papillae, circular amphids, a strongly annulated cuticle, a narrow tubular buccal tube, a narrow cervical region expanding to accommodate the strongly muscular pharynx, two outstretched ovaries ventral to the gut and simple curved spicules. The taxonomic placement of the Peresianidae is difficult but the conclusion of other taxonomists that it belongs within the Leptolaimina is supported.

KEY WORDS: Taxonomy, marine nematodes, Peresianidae, *Manunema*.

Introduction

Manunema pectenophora sp. nov. possesses prominent male supplementary organs, i.e. ventral pre-anal organs found in many male nematodes, but in the new species they are unlike those described previously. The Peresianidae contains a single genus, *Manunema*, comprising only two previously-described species, namely *M. proboscidis* Gerlach, 1957, and *M. annulata* (Vitiello & de Coninck 1968) Riemann, *et al.* 1971.

The taxonomic placement of the Peresianidae has proved a problem. Some characters suggest placing the family in the Leptolaimina (Chromadorida), others are closer to the Desmoscolecoidea (Monhysterida).

Materials and Methods

Specimens were collected from the intertidal zone of beaches at Darwin NT. Samples of about 2 kg of sand were dug up at low tide and the meiofauna present was briefly suspended in 5 litres of tap water with vigorous stirring. As soon as the sand had settled, the water was passed through a 60 μm nylon sieve and the fauna retained on the sieve back-washed into a beaker with sea water. They were immediately fixed by adding formalin to give a final concentration of 5%. Later, the meiofauna was examined in petri dishes under a binocular microscope. The new species was isolated by pipette from the many hundreds of other nematodes collected and the nematodes mounted on microscope slides in anhydrous glycerol. Cover slips were supported by glass beads (Ballatini) selected under the microscope to be slightly wider than the nematodes and the cover slips were ringed with Glyceel (Gurr).

Measurements are in μm from specimens fixed and mounted in this way. De Man's indexes (ratios) (Fortuner 1990) are given, i.e. a = body length divided by greatest body width, b = length divided by length of pharynx, c = length divided by tail length, c' = tail length divided by width at anus, V = anterior end to vulva as a percentage of body length, and spicule measurements are arc length.

Drawings and measurements were made using a camera lucida. When mounted, the nematodes lie on their sides presenting a lateral view, and our drawings, with the exception of all four cephalic setae, show setae on one side only, those lying uppermost as mounted.

For scanning electron microscopy, some specimens in 5% formalin were washed in phosphate buffer, pH 7, containing 3% sucrose, post-fixed by the addition of 2% osmium tetroxide, washed, sonicated and finally freeze-dried. The specimens were mounted on metal stubs and coated with gold/palladium before examination in the microscope.

Type specimens are deposited in The South Australian Museum, SAMA, Adelaide, and their numbers in the Museum's Australian Helminth Collection, AHC, are given in the text.

Manunema pectenophora sp. nov. (FIGS 1-11)

Holotype: Male, Rapid Creek beach, Darwin, NT, 19.x.1992, SAMA, AHC 30000.

Measurements: Table 1

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TABLE 1. *Measurements of* Manunema pectenophora *sp. nov.*

Type	Holo Male	Male paratypes n = 3 Range	Mean	±SD	Female paratypes n = 4 Range	Mean	±SD
Length	488	468-506	490	20	475-508	497	17
Maximum width	15	11-12	13	0.58	16-20	18	1.83
Cephalic setae	13	10-16	13	3.06	12-18	15	2.50
Body setae	11	11-14	12	1.73	10-14	12	1.83
Mouth to amphid	12	10-11	10	0.58	9-9	9	0.50
Amphid diameter	3.7	3.0-3.2	3.1	0.12	3.6-3.6	3.6	0.00
Width at amphid	6.5	5.0-6.5	5.8	0.76	5.6-5.6	5.6	0.05
Buccal cavity	30	32-36	34	2.08	31-34	33	1.50
Width at buccal cavity	12	9-11	10	1.15	9-12	11	1.50
Mouth to nerve ring	57	54-60	57	3.06	55-60	58	2.08
Width at nerve ring	13	13-14	13	0.58	14-18	16	1.73
Pharynx	83	77-83	80	3	82-88	85	2.58
Width at cardia	14	14-17	14	2.52	10-18	14	2.99
Mouth to vulva	-	-	-	-	252-273	265	10
Width at vulva	-	-	-	-	15-20	18	1.83
Egg	-	-	-	-	44-79	57	20
Mouth to anus	341	391-434	412	22	407-438	426	14
Tail	81	72-84	78	6	67-74	71	3.77
Width at anus	11	11-11	11	0	8-11	10	1.29
Spicule, arc length	24	23-25	24	1			
Gubernaculum	11	10-11	10	0.99	-	-	-
Anus to 1st supplement*	6.8	4.5-7.3	6.4	1.57	-		-
Anus to 2nd supplement*	13	11-15	13	2.25	-		-
Anus to 3rd supplement*	34	31-34	33	1.62	-	-	-
De Man's a	33	36-39	39	2.63	25-30	28	2.6
De Man's b	5.9	5.6-6.6	6.1	0.47	5.7-6.2	5.8	0.24
De Man's c	6	5.9-7.0	6.3	0.63	6.9-7.3	7.0	0.22
De Man's c'	7.4	6.5-7.6	7.1	0.55	6.7-8.5	7.5	0.96
De Man's V%	-	-	-	-	51-55	53	1.48

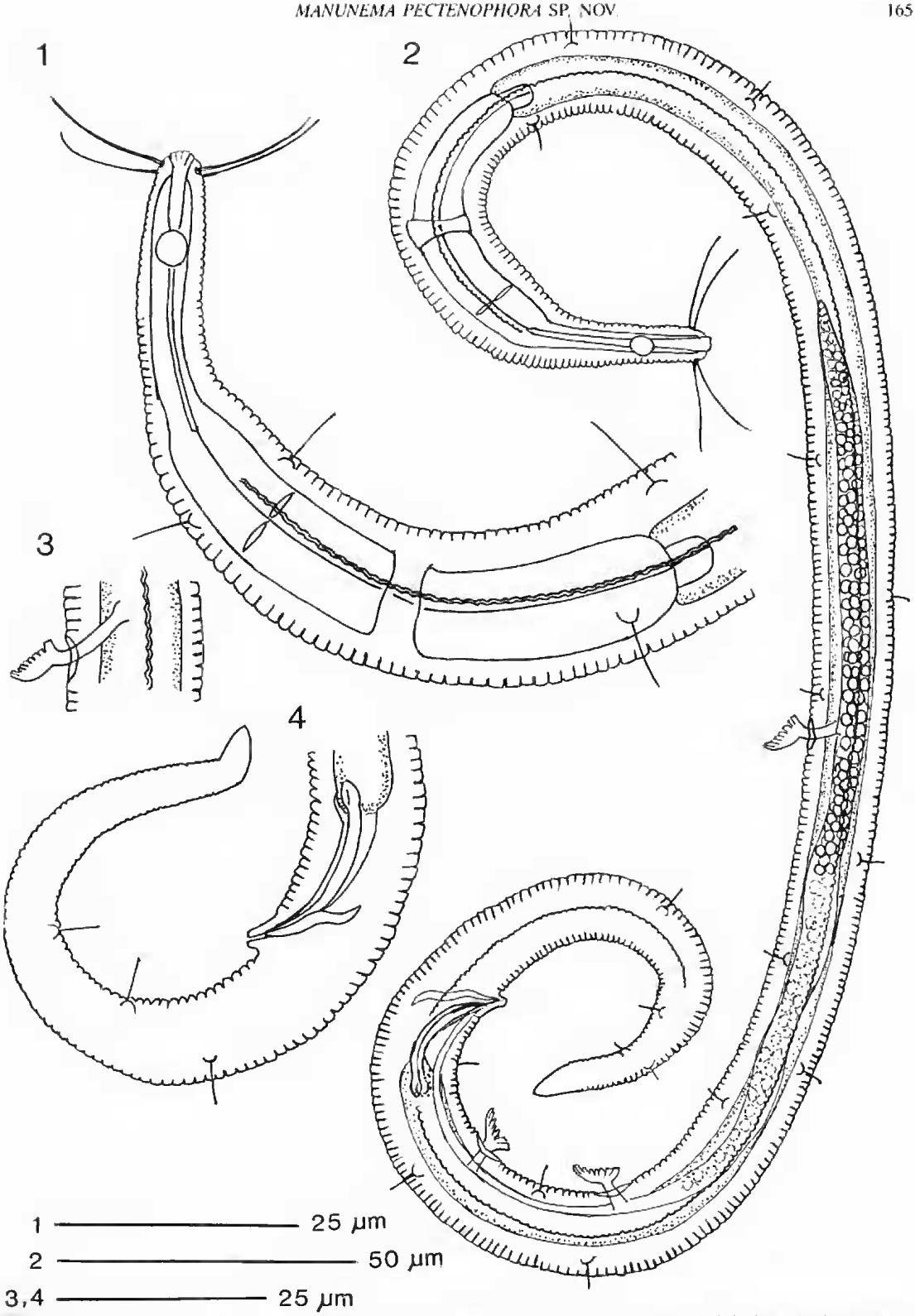
*As percentage of body length

Description of Holotype male

Small, body when fixed strongly curved, head and cervical region folded back along body, tail curled. Cuticle strongly annulated; lateral ridges from mid-pharyngeal region to mid tail, wavy in register with annules; four rows of prominent body setae, arising from pronounced cuticular hemispherical swellings, dorso-lateral setae alternate with ventro-lateral setae. Four long cephalic setae arising form sockets; labial setae absent; amphid circular. Buccal cavity, with minute ridges around mouth, initially narrowly conical extending posteriorly as a narrow parallel-sided tube. Pharynx, in cervical region (35% of pharynx length) narrow parallel-sided, encloses buccal tube, then a wider muscular cylinder, somewhat constricted by prominent nerve ring, two cytoplasmic clefts between nerve ring and expansion; cardia short, cylindrical. Intestine simple tube, anus and rectum project slightly from body contour; caudal glands not observed (probably obscured by strong annulation). Single testis to left of intestine; spicules cephalated, smoothly curved, tips pointed; gubernaculum slightly curved plate. Three pre-anal supplementary organs, most anterior one about mid-way between cardia and anus, the other two close to anus. Each supplement resembles an outwardly and slightly forwardly directed comb, with about 11 prongs, mounted on a cuticular rod arising deep in the body wall.

Paratypes: SAMA. AHC 30001-7. Measurements of three males and four females are given in Table 1. In paratype males, as in the holotype, anterior supplement about 33% of body length in front of anus, second and third supplements, closer to anus, apparently more variable in position, probably due to different degrees of body curvature. Long testis, to left of intestine, with many developing sperm, begins just anterior to mid body, continues as long sperm duct. SEM of another male, Figs 6 and 7, shows a tenous transparent film overlapping the base of a supplement and adjacent cuticle. We interpret this as mucus, present over the surface of freshly fixed specimens and preserved by freeze-drying but lost when specimens are transferred to glycerol for light microcopy.

Females (Fig. 5) similar to males apart from reproductive organs and absence of supplementary organs. Didelphic, two very short ovaries outstretched, ventral to gut. Three females each have single large egg, 43, 48 and 79 μm long, respectively, overlapping the vulva, to left of intestine. The largest is probably at an early stage of the first cleavage division.



Figs 1–4. *Manunema pectenophora* sp. nov. 1. Male head. 2. Entire male. 3. Supplementary organ. 4. Spicules and gubernaculum.

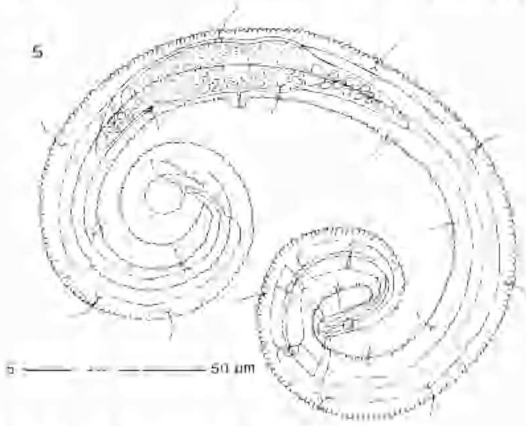


Fig. 5. Female *Manunema pectenophora* sp. nov.

Differential diagnosis

The form of the supplement distinguishes *M. pectenophora* sp. nov. from the other described species of *Manunema*, none of which possesses comb-like structures mounted on rods. The new species differs from *M. annulata* in the orientation of the single testis.

Habitat

Sandy ocean beach.

Distribution

So far known only from Rapid Creek beach, a suburb of Darwin, Northern Territory.

Etymology

Named from L. *pecten*, a comb.

Discussion

Supplementary organs are common in many families of Adenophorea, where they are associated with sensilla, and are generally believed to play a part in copulation. They may be tubular, setose or papilliform and are often associated with cuticular ornamentation but none like the organs described here has previously been reported. They do not appear to be associated with sensilla and conceivably serve some mechanical role in copulation. *M. proboscidis* possesses two pre-anal tubular male supplementary organs (Gerlach 1957). Vitiello & de Coninck (1968) claimed that supplements were lacking in *M. annulata*, but Riemann *et al.* (1971) redescribed *M. annulata*, reporting two pre-anal tubular supplements. Neither Gerlach (1957) nor Vitiello & de Coninck (1968) comment on the buccal cavity. We agree with Riemann *et al.* (1971) that the buccal cavity is long and tubular. Lorenzen (1981) includes a long tubular buccal cavity as one of the diagnostic characters of the Peresianidae.

The taxonomic position of the Peresianidae, to which *Manunema* belongs, has been the subject of some doubt, partly because their small size has led to some uncertainty about taxonomically important characters. It is significant that scanning electron microscopy does not show either outer labial papillae or setae, nor any external manifestation of inner labial sensilla. All the described species have four long sub-median cephalic setae inserted in sockets.

In Lorenzen's (1981) phylogenetic classification of the Adenophorea, ovaries ventral to the intestine and a single posterior testis are significant characters in *Manunema*, consistent with the placement of the Peresianidae in the Leptolaimina but, while the location and form of the ovaries in *M. pectenophora* are the same as in *M. proboscidis*, we have observed a single anterior testis in three males of *M. pectenophora*. The form of the amphids, the long narrow buccal tube and tubular supplementary organs are consistent with Leptolaimina; ventral outstretched ovaries are not (Lorenzen 1981). In the possession of four cephalic setae, the absence of outer labial setae, the possession of four sub-median rows of alternating body setae arising from peduncles and the anus on a protrusion from the body cavity, *Manunema* resembles the Desmoscolecoidea, within the Monhysterida, rather than the Leptolaimina.

In Vitiello and de Coninck's (1968) view, the similarities between *Peresiana annulata*, now renamed *Manunema annulata* Riemann *et al.* (1971), and *Meylia spinosa* Gerlach 1956 indicated a phylogenetic link between the Haliplectidae (Leptolaimina in Lorenzen's classification) and the Desmoscolecida, in which they placed the new species. The similarities to which they drew attention were the four cephalic setae and the position of the non-vesicular amphids, but in other respects the species are unalike, differing in the structure of the cuticle, buccal cavity, pedunculate setae and the location of the anus. In fact, as Riemann *et al.* (1971) point out, there are similarities between *Manunema* and other Desmoscolecoidea, for example with *Tricoma mirabilis* Timm 1961, although *Manunema* shows greater similarity with such Leptolaimina as *Anomonema haplostoma* Hopper 1963 and *Leptolaimus tritubulatus* Boucher and Helléouët 1977.

Although *M. pectenophora* does not possess tubular supplementary organs or a posterior testis (leptolaimid characters of *Manunema proboscidis* and *M. annulata*) we concur with the placement of Peresianidae in the Leptolaimina, with a possible link between Leptolaimina and Desmoscolecoidea.

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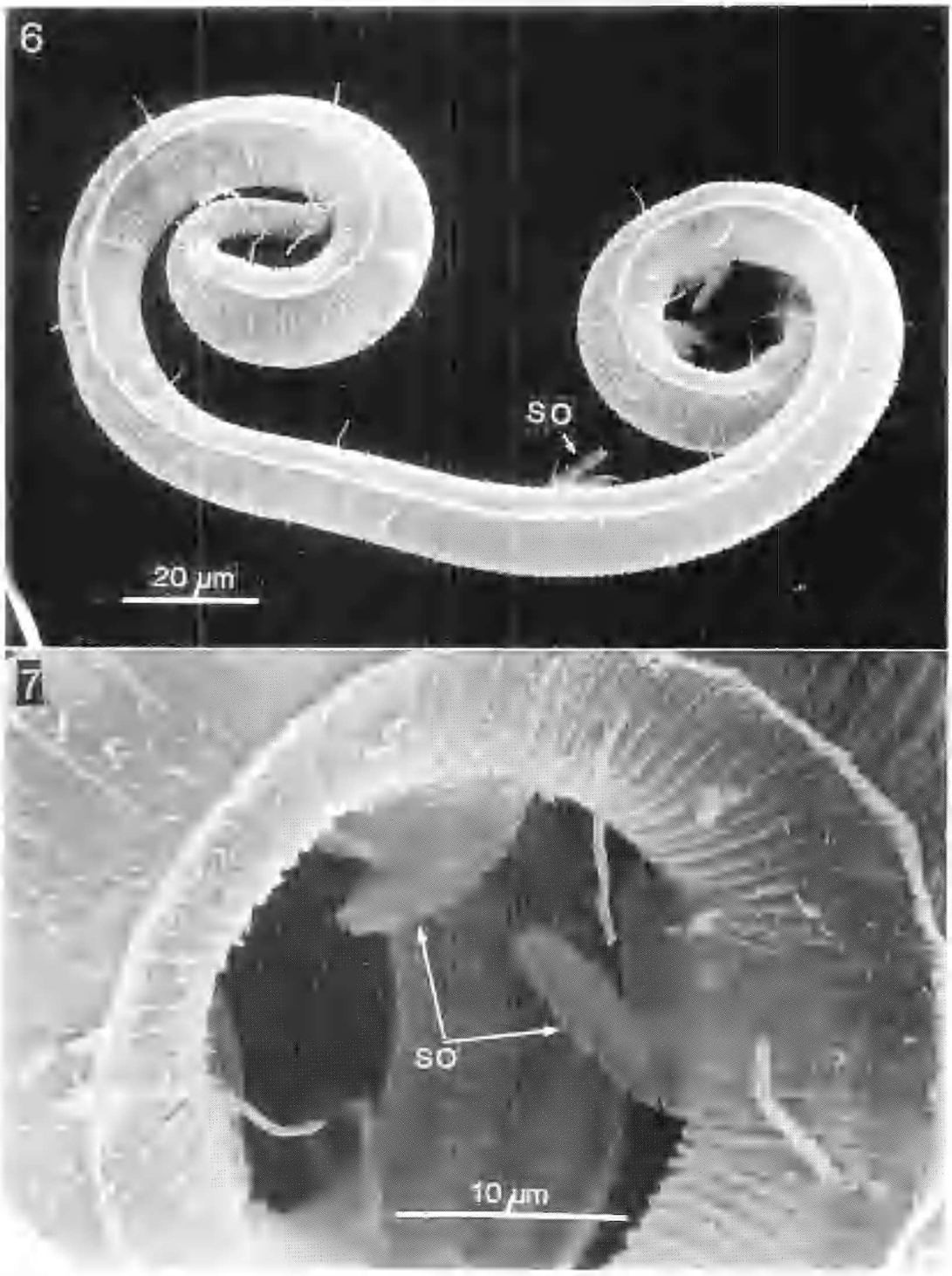
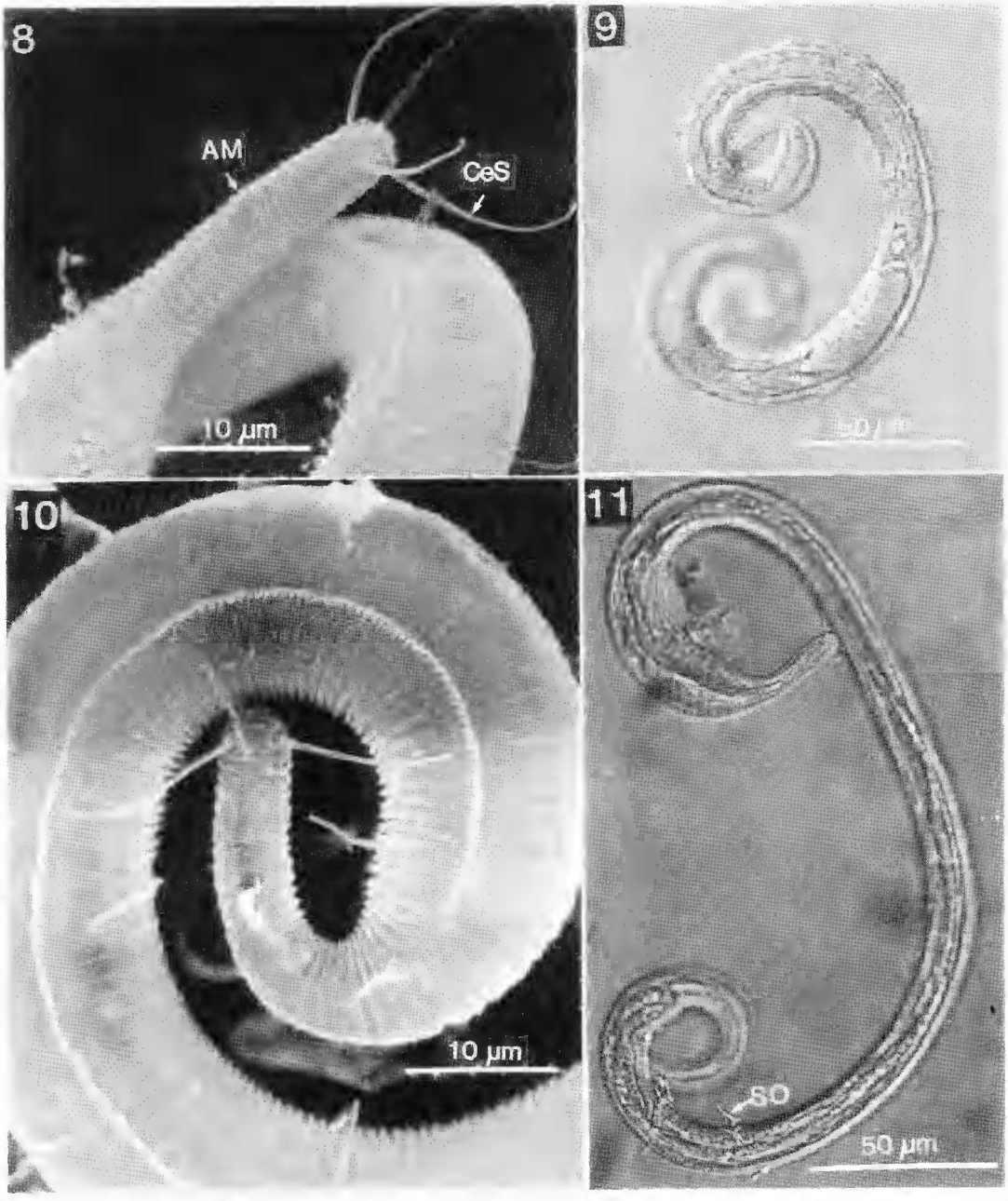


Figs 6 and 7. Scanning electron microscopy of *Manunema pectenophora* sp. nov. 6. Entire male. 7. Enlargement to show supplementary organs. SO supplementary organ.



Figs 8-11. 8. Scanning electron microscopy of female *Manunema pectenophora* sp. nov. CeS cephalic seta, AM amphid. 9. Female by light microscopy. 10. SEM of female head. 11. Male by light microscopy. SO supplementary organ.

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